

**PATENT CLAIMS**

1. Polynucleotide which codes for a protein with trans-sialidase activity and can be isolated from *Trypanosoma congolense*.

2. Polynucleotide in accordance with claim 1, which codes for a protein with trans-sialidase activity

and which catalyses the transfer of sialic acid from a donor onto an acceptor molecule.

3. Polynucleotide in accordance with claim 1 or 2, comprising a nucleic acid sequence in accordance

with SEQ ID NO: 1 or 3, or fragments of the same, which comprise at least 15 connected nucleotide residues; the polynucleotides and fragments complementary to the same; and the nucleotide sequences derived from these polynucleotides by degeneration of the genetic code.

4. Oligonucleotide which hybridises with a polynucleotide in accordance with one of the previous

claims, in particular under stringent conditions.

5. Polynucleotide which hybridises with an oligonucleotide in accordance with claim 4, in particular

under stringent conditions, and codes for a gene product of microorganisms of the *Trypanosoma* genus.

6. Polypeptide which is coded by a polynucleotide which comprises a nucleic acid sequence in accordance with any of the claims 1 to 3 or 5; or which has an amino acid sequence which comprises at least 10 connected amino acids in accordance with SEQ ID NO: 2 or 4; and functional equivalents of the same which have trans-sialidase activity.

7. Trans-sialidase or functional equivalents of the same with trans-sialidase activity, characterised by

one of the following amino acid part sequences:

TDTVKYSTDGGRTWKREVIIPNGR (pos. 1 to 25 in accordance with SEQ ID NO: 2)

FRIPSLVEIDGVLIAFDTRYLRAASDSSLI (pos. 1 to 30 in accordance with SEQ ID NO: 4).

8. Trans-sialidase 1 (TS1) characterised by at least one of the following characteristics:

Nucleotide sequence	SEQ ID NO: 1
Amino acid sequence	SEQ ID NO: 2
Temperature optimum	30-40°C
pH optimum	pH 6.5-8.5
Isoelectric point	pH 4-5
Molecular weight, native	400-600 kDa
Molecular weight in	90 kDa
the reducing SDS page	

9. Trans-sialidase 2 (TS2), characterised by at least one of the following characteristics:

Nucleotide sequence	SEQ ID NO: 3
Amino acid sequence	SEQ ID NO: 4
Temperature optimum	30-40°C
pH optimum	pH 6.5-8.5
Isoelectric point	pH 5-6
Molecular weight, native	120-180 kDa
Molecular weight in the	
reducing SDS page	90 kDa

10. Material in accordance with any of the claims 1 to 9, derived from the *Trypanosoma congoense*

organism.

11. Materials in accordance with any of the claims 1 to 9, produced using synthetic, in particular

chemical, biochemical, enzymatic, gene technological and transgenic methods.

12. Functional equivalent of a trans-sialidase in accordance with either of the claims 8 and 9, the amino acid sequence or part sequence of which has a sequence sameness of at least 50 % or at least 60 %, in particular at least 65 % or at least 70 %, such as eg. 75 %, 80%, 85 %, 90 %, 95 %, 98 % or 99% to the corresponding amino acid sequence or part sequence in accordance with SEQ ID NO: 2 or 4, calculated according to the algorithm of Pearson and Lipman, Proc. Natl. Acad. Sci. (USA) 85(8), 1988, 2444-2448; or which contains one or more deletions, additions, substitutions or inversions of an individual or of several amino acid residues or shows a changed glycosylation pattern; whereby the capability of catalysis of the transfer of sialic acids from a donor to an acceptor is maintained.
13. Expression cassette, comprising, in operative connection with at least one regulative nucleic acid sequence, a nucleic acid sequence in accordance with any of the claims 1 to 5.
14. Recombinant vector, comprising at least one expression cassette in accordance with claim 13.
15. Prokaryotic or eucaryotic host, transformed with at least one vector in accordance with claim 14.
16. Use of an expression cassette in accordance with claim 13, of a vector in accordance with claim 14 or of a host in accordance with claim 15 for the recombinant production of a protein with trans-sialidase activity.
17. Method for the enzymatic sialylation of an acceptor molecule, characterised in that the acceptor molecule is incubated with a donor containing sialic acid residues in the presence of an enzyme in accordance with any of the claims 6 to 12, and the sialated acceptor is isolated.

18. Method in accordance with claim 17, characterised by at least one more of the following properties:

- a) the donor is chosen from sialic acids bonded to oligosaccharides, polysaccharides, polysialic acids, glycoproteins and glycolipids, such as in particular lactoferrins, glycolysated whey proteins and caseins, and fragments of the same;
- b) the acceptor is chosen from polymers containing  $\beta$ -galactose, such as  $\beta$ -galactooligosaccharides, lactitol, lactobionic acid, methyl- $\beta$ -lactoside, acetyllactosamines, galactopyranosides, trans-galactooligosaccharides, polygalactose and other glycoconjugates with terminally bonded  $\beta$ (1-3) or  $\beta$ (1-4) galactose or galactose.

19. Use of a trans-sialidase in accordance with any of the claims 6 to 12, of a nucleic acid sequence

coding for the same in accordance with any of the claims 1 to 5 or of a sialidase product produced in accordance with either of the claims 18 and 19 for the production of a medicament, foodstuff or food additive, or of a food stuff additive for the prevention or treatment of parasitic, bacterial or viral infections controlled by sialic acid; for the treatment of tumour diseases; for the treatment of diseases which are associated with development interference of the tissue; for the treatment of diseases of the immune system; for the treatment of autoimmune reactions; for the treatment of diseases with interrupted cell communication; or for the treatment of inflammation.

20. Use of a trans-sialidase in accordance with any of the claims 6 to 12, for the development of a Trypanosomiasis vaccine or for the development of enzyme inhibitors for the treatment or prevention of Trypanosoma infections.

21. Use of a trans-sialidase in accordance with any of the claims 6 to 12, of a nucleic acid sequence

coding for the same in accordance with any of the claims 1 to 5 or of a sialated product produced in accordance with either of the claims 18 and 19 for the production of a medicament, food additive or

food stuff for the protection of the body's own cells or tissues or glycoproteins before the enzymatic effect.

22. Use of a trans-sialidase in accordance with any of the claims 6 to 12, of a nucleic acid sequence

coding for the same in accordance with any of the claims 1 to 5, or of a sialated product produced in accordance with either of the claims 17 and 18 for the production of a medicament, food additive or food stuff in order to influence the development and/or morphogenesis of body tissues.

23. Effector of the trans-sialidase activity of a trans-sialidase in accordance with any of the claims 6 to

12, chosen from

a) polypeptide ligands which interact with a trans-sialidase in accordance with any of the claims 6 to 12;

b) low molecular effectors which modulate the biological activity of a trans-sialidase in accordance

with any of the claims 6 to 12; and

c) antisense nucleic acid sequences of a nucleic acid sequence in accordance with any of the claims 1

to 5.

24. Use of an effector in accordance with claim 23 for the production of a pharmaceutical or gene-therapeutic means, food additive or foodstuff for the treatment or prevention of diseases associated with trans-sialidase activity.

25. Method for the isolation of an enzyme with trans-sialidase activity, whereby

a) *Trypanosoma congolense* is cultivated in a medium,

b) and the desired product is isolated from the culture supernatant by means of *ion exchange*

*chromatography* with the help of a salt gradient, if required followed by isoelectric focussing, gel filtration, affinity chromatography and/or protein precipitation.

26. Pharmaceutical or gene-therapeutical means, containing in a pharmaceutically or gene-therapeutically compatible carrier at least one effector in accordance with claim 23.